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# Effects of substrate size and cleaning regime on growth and survival of captive-bred juvenile freshwater pearl mussels, *Margaritifera margaritifera* (Linnaeus, 1758)

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**2nd International Seminar on Rearing of Unionoid Mussels**  
**24-27 November 2015, Clervaux, Luxembourg**



# What we'll cover

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- Experiment 1 – Effect of substrate size and cleaning regime on survival and size
- Experiment 2 – Investigating environmental factors affecting survival and size
  - Flow rate
  - Interstitial space
  - Dissolved oxygen
  - Ammonia concentration

# Initial experiment – Exp 1

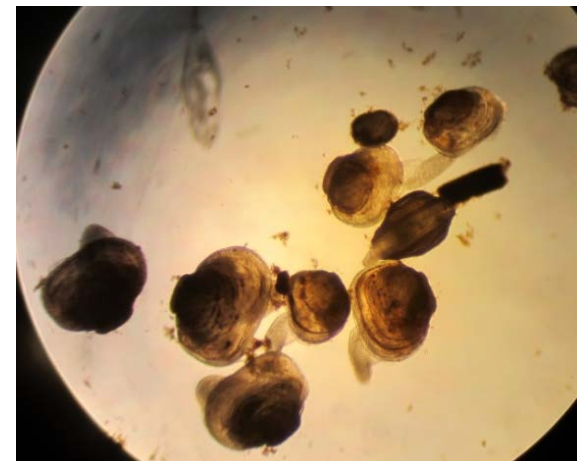
- In 2012, 3600 juveniles (9 replicates) divided into 4 treatments:
  - 0.25-1 mm cleaned weekly
  - 0.25-1 mm cleaned monthly
  - 1-2 mm cleaned weekly
  - 1-2 mm cleaned monthly



## Exp 1 - Method

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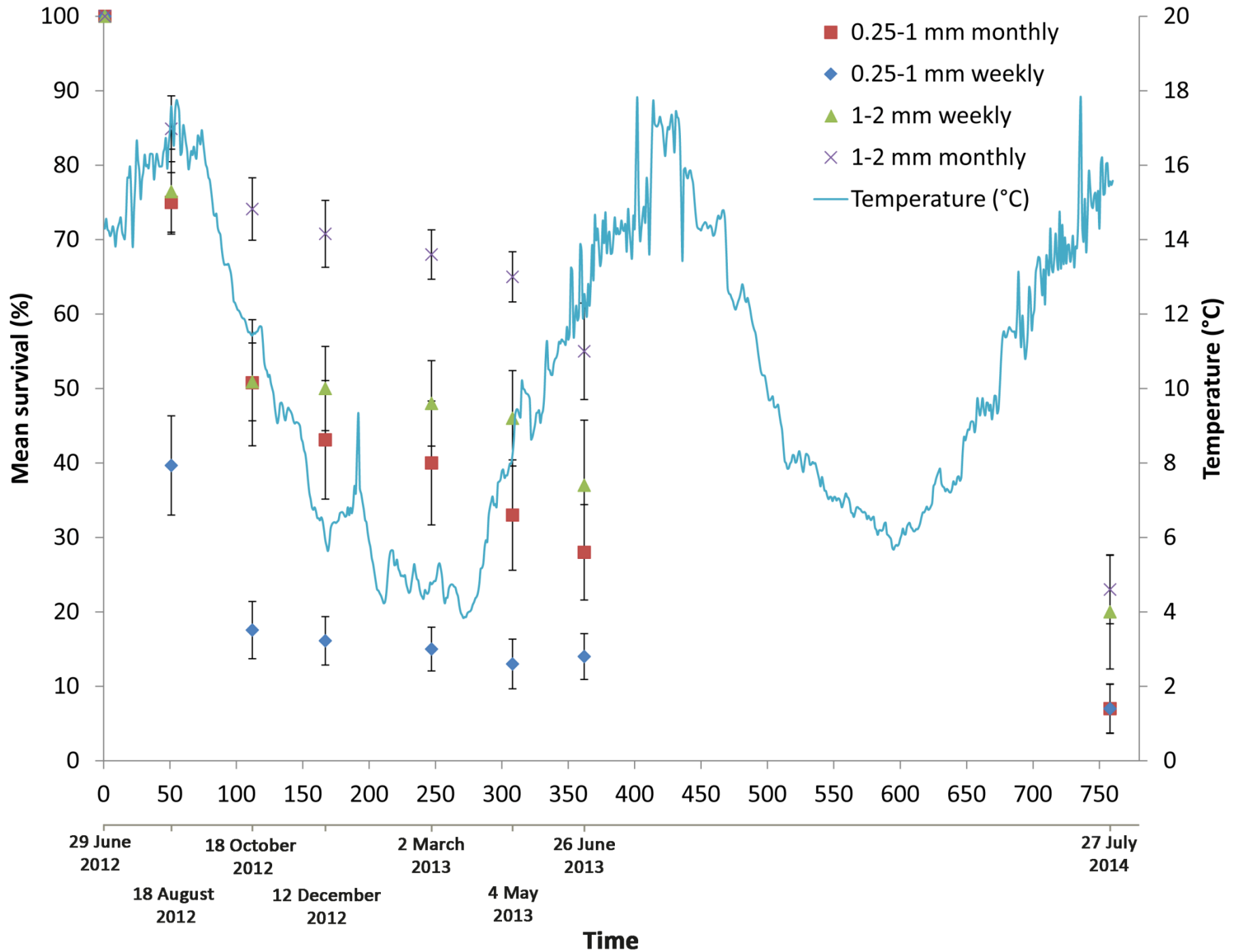
- Cleaning regime maintained for 25 months
- Substrate emptied into a glass dish, elutriated and poured through a 0.18 mm sieve to catch juveniles
- Sampled every 2 months for growth and survival during first 12 months, then final check at 25 months



# Exp 1 - Results

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- Survival



# Exp 1 - Results

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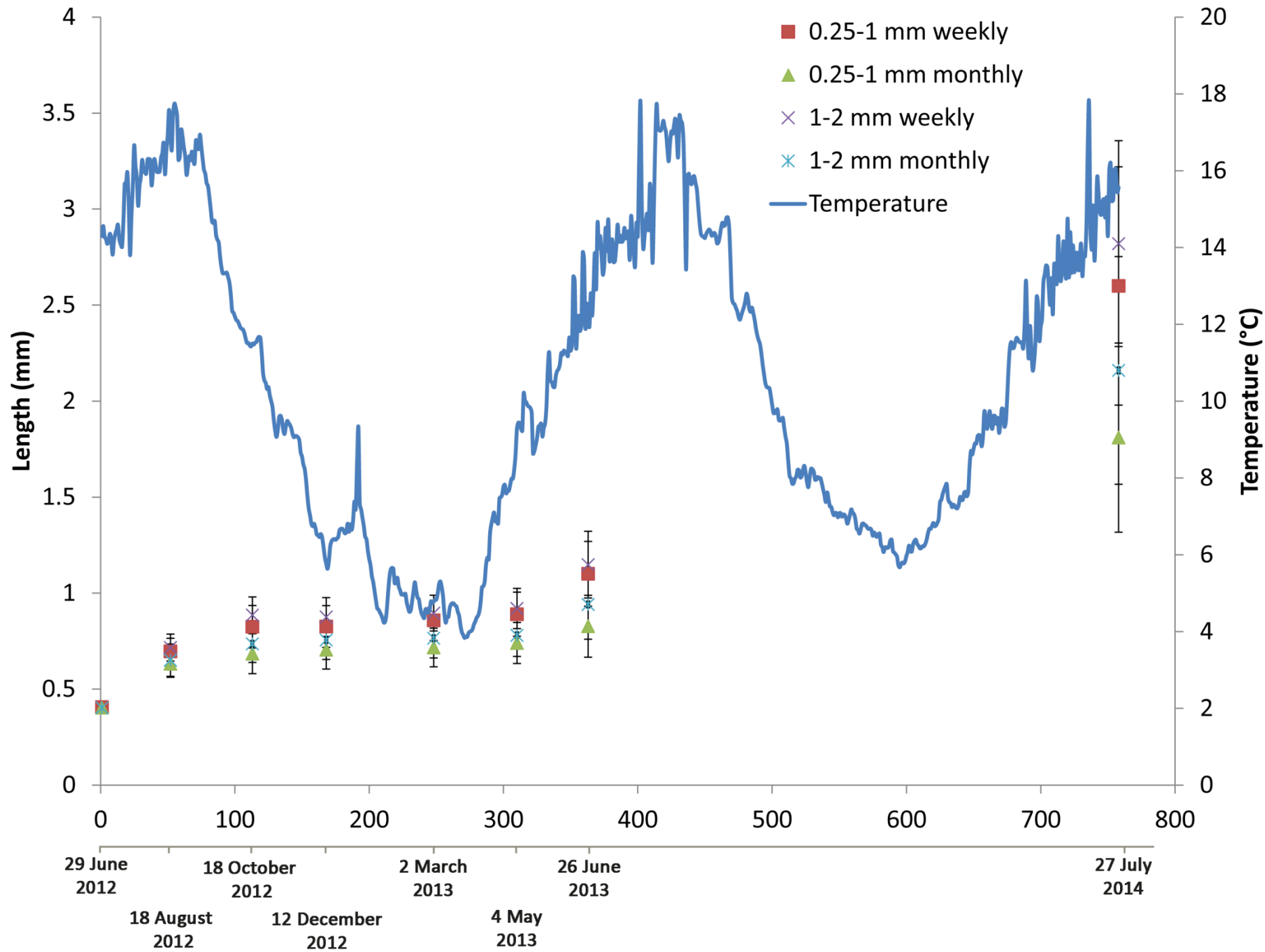
- Survival
  - Significantly ( $P < 0.001$ ) different on all sampling occasions
  - On day 362 all treatments were significantly different from each other but by day 758 two groupings became obvious, larger and smaller substrate
  - 1-2 mm monthly > 1-2 mm weekly > 0.25-1 mm monthly > 0.25-1 mm weekly
  - Mortality slowed over winter but did not stop completely



# Exp 1 - Results

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- Size



# Exp 1 - Results

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- Size
  - Significantly different ( $P < 0.001$ ) on all sampling occasions
  - Weekly treatments were statistically the same on day 362 ( $P = 0.058$ ) but all treatments were significantly different by day 758 ( $P < 0.001$ ) and size ranges increase
  - 1-2 mm weekly > 0.25-1 mm weekly > 1-2 mm monthly > 0.25-1 mm monthly

## Exp 1 – Conclusions

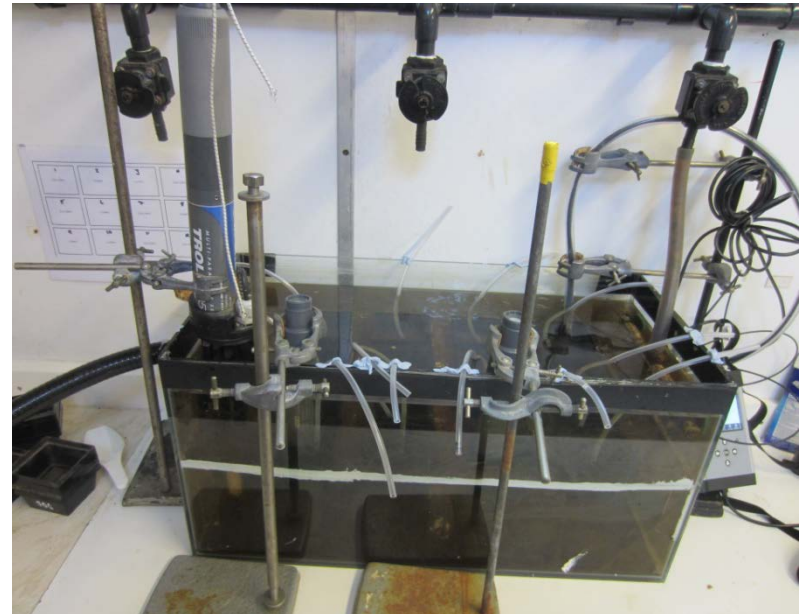
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- Higher survival in larger substrates
- Higher growth in substrates cleaned weekly
- Near cessation of growth  $< 10^{\circ}\text{C}$  concurring with previous findings e.g. Ziuganov *et al.*, 1994; Buddensiek, 1995; Hruška, 1999
- No size-dependent over-winter survival observed in any treatment
- *...but why these differences???*

## Additional experiment – Exp 2

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- Investigating factors which may affect growth and survival:
  - Flow rate through substrate & interstitial space
  - Dissolved oxygen
  - Ammonia concentration at 4 weeks



## Exp 2 - Methods

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- Same experimental set up but only 3 replicates
- Took place over 8 weeks starting 21/07/2015

### 1. Flow rate:

- Measured time to clear 1L of water
- Repeated after substrate had been cleaned

### 2. Interstitial space:

- Emptied substrate into measuring cylinder and topped up with water until meniscus touched top of substrate

## Exp 2 - Methods

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3. DO – logged every 15 minutes over weeks 5-8
  - **Water column** - multi-parameter sonde
  - **Monthly treatments** – Onset DO loggers
  - **Weekly treatments** – PreSens DO dipping probe
4. Ammonia concentration of interstitial water taken after 4 weeks by siphoning 2 ml out of sieves using plastic tubing (spectrophotometer)



## Exp 2 - Results

- Survival (%)

Treatment	2012	2015
0.25-1 mm weekly	40 $\pm$ 7	72 $\pm$ 2
0.25-1 mm monthly	75 $\pm$ 4	68 $\pm$ 4
1-2 mm weekly	76 $\pm$ 6	80 $\pm$ 4
1-2 mm monthly	85 $\pm$ 4	81 $\pm$ 8



## Exp 2 - Results

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- Juvenile length (mm) was not significantly different in 2015 ( $P = 0.53$ )

Treatment	2012	2015
0.25-1 mm weekly	0.72 $\pm$ 0.07	0.82 $\pm$ 0.10
0.25-1 mm monthly	0.70 $\pm$ 0.07	0.83 $\pm$ 0.08
1-2 mm weekly	0.65 $\pm$ 0.08	0.84 $\pm$ 0.10
1-2 mm monthly	0.63 $\pm$ 0.07	0.81 $\pm$ 0.09

- More analysis required on size differences & relationship to temperature

## Exp 2 - Results

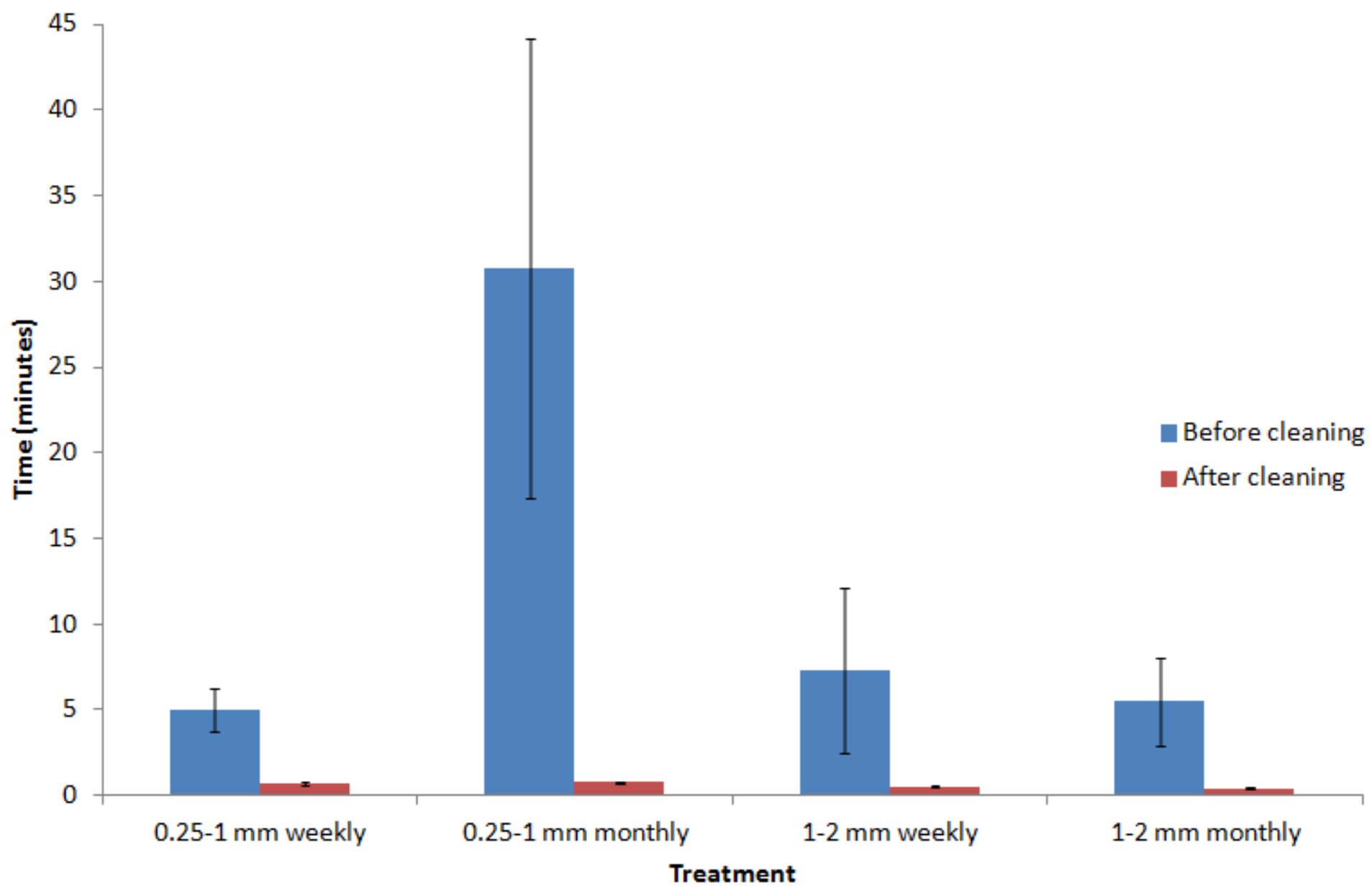
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### 1. Flow rate:

- Pre-cleaning, flow through 0.25-1 mm monthly treatment was significantly slower than all other treatments ( $F_{(3,8)} = 8.83$ ,  $P = 0.006$ )
- Flow through cleaned substrates was significantly higher ( $F_{(3,8)} = 18.80$ ,  $P = 0.001$ ) in the 1-2 mm compared to the 0.25-1 mm substrates

### 2. Interstitial space:

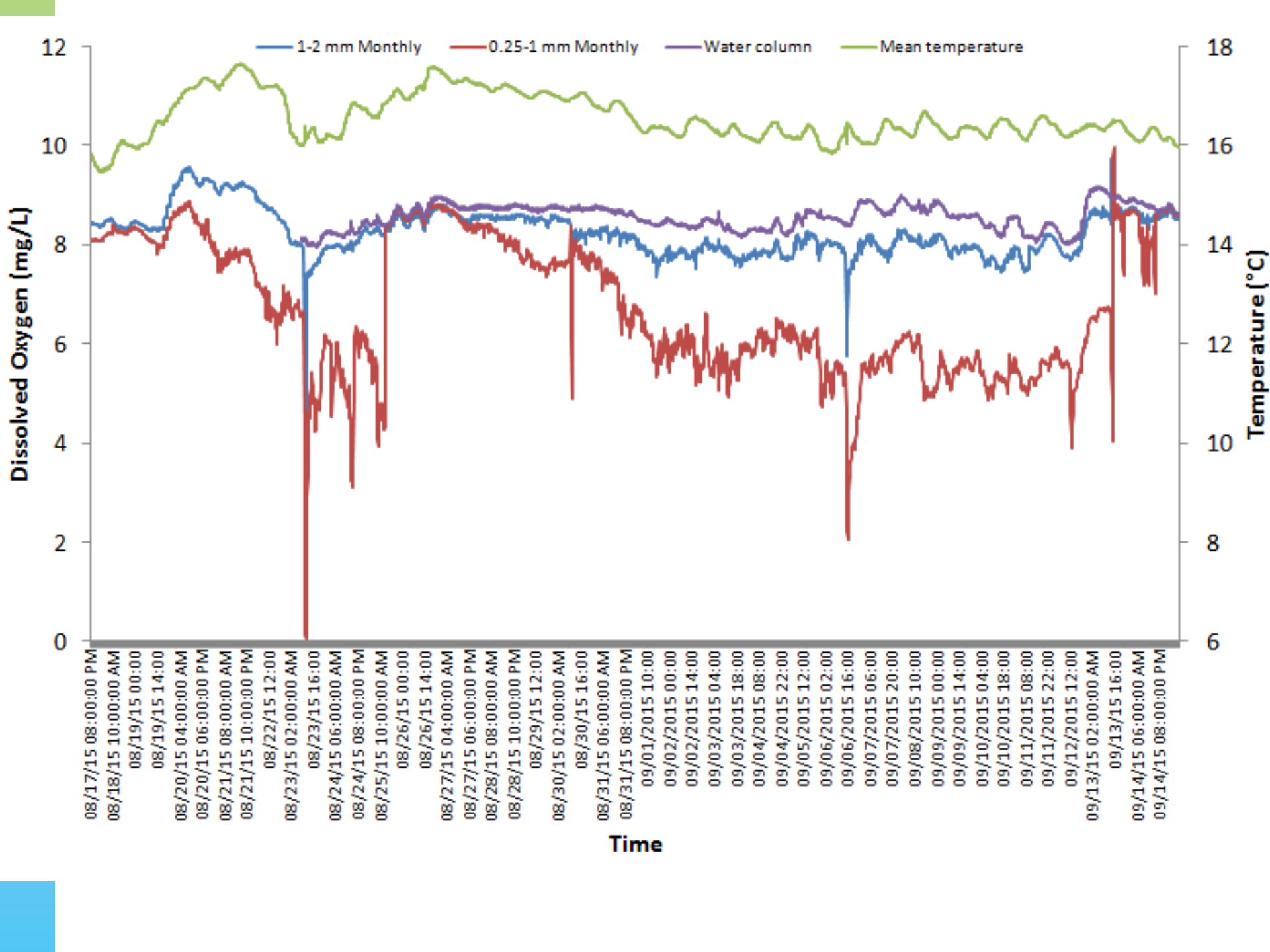
- 0.25-1 mm substrates had significantly less ( $t_{(10)} = -4.72$ ,  $P = 0.001$ ) interstitial space ( $2.6 \pm 0.61$  ml) compared with 1-2 mm substrates ( $4.3 \pm 0.25$  ml)

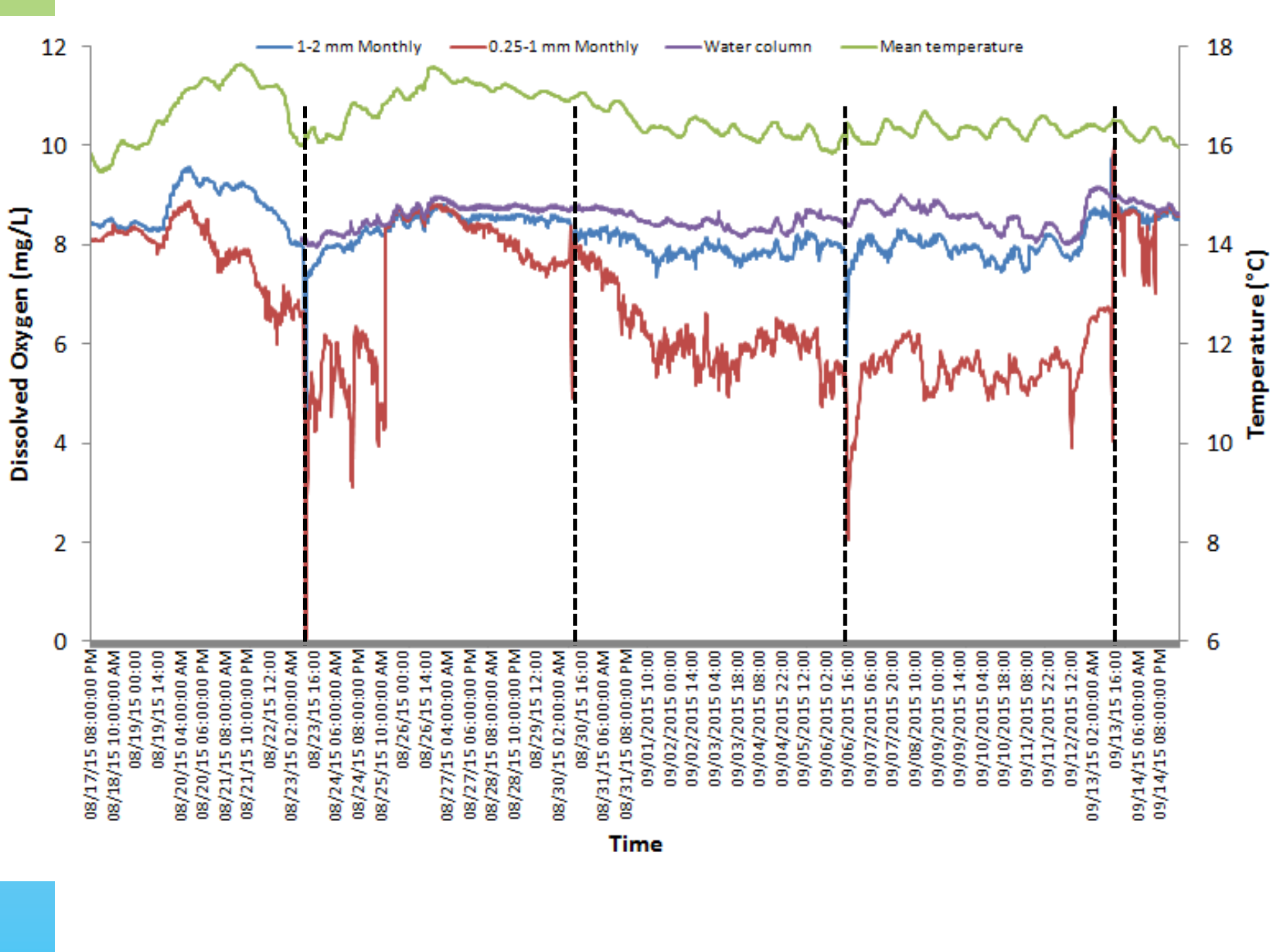


# Exp 2 – Results

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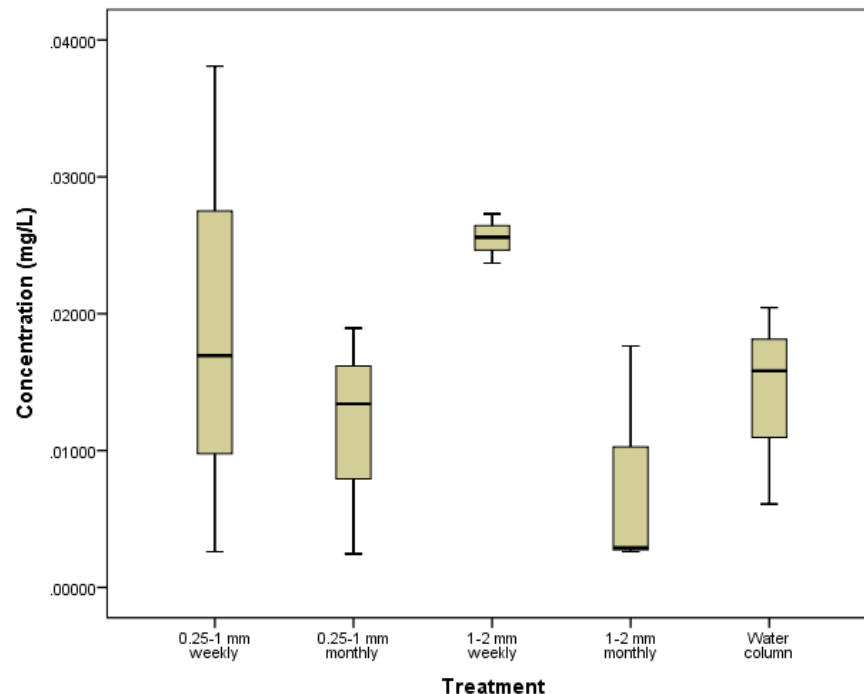
## 3. Dissolved Oxygen





## Exp 2 - Results

- Ammonia concentrations were the same between the four treatments ( $F_{(4, 14)}=1.38$ ;  $P=0.307$ )



# Conclusions

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- Larger substrate provides better survival rates
- Growth patterns were different between exp 1 & 2
- DO and flow are significantly affected by substrate size and cleaning regime
- Further analysis of DO data will help explain this further



# References & acknowledgements

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- Buddensiek, V. (1995) The culture of juvenile freshwater pearl mussels *Margaritifera margaritifera* L. in cages: A contribution to conservation programmes and the knowledge of habitat requirements. *Biological Conservation* **74**: 33-40.
- Hruška, J. (1992) The freshwater pearl mussel in South Bohemia: Evaluation of the effect of temperature on reproduction, growth and age structure of the population. *Archiv für Hydrobiologie* **126**: 181-191.
- Ziuganov, V., Zontin, A., Nezlin, L. and Tretiakov, V. (1994) *The freshwater pearl mussels and their relationships with salmonid fish*. VNIRO Publishing House, Moscow.

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